Hypersensitivity pneumonitis in a *Pleurotus* mushroom grower

Konstantinos Loverdos, Iris Karahaliou, Erasmia Economidou, Mina Gaga, Likurgos Kolilekas

7th Pulmonary Department and Asthma Center, Athens Chest Hospital, Greece

Abstract. A 47 year old woman presented to the emergency department due to persistent productive cough, accompanied by breathlessness on exertion, chest pain and weight loss during the last three months. She was an occasional smoker and had been working as a mushroom grower of the *Pleurotus* species during the past six months, with an otherwise unremarkable medical history. An earlier chest CT scan, performed forty days previously, revealed bilateral patchy ground glass opacities, more profound in the upper lobes. One month before presentation she underwent bronchoscopy in another hospital and the cytological examination of the collected bronchoalveolar lavage (BAL) demonstrated excessive neutrophilia. She received at that time a course of antibiotic therapy with doxycyclin and amoxicillin/clavulanate, without clinical improvement. During hospitalization in our department, the patient underwent again bronchoscopy with BAL, revealing significant lymphocytosis (32%), additionally to the previously observed neutrophilia (43%). Given the compatible occupational history, the radiologic pattern and the BAL subpopulation analysis, a diagnosis of hypersensitivity pneumonitis was made and the patient was discharged with a recommendation to withdraw from her current occupational activity. Six weeks later, the patient presented evident clinical, imaging and functional improvement. Pneumon 2015, 28(2):179-184.

Key words: - Hypersensitivity pneumonitis - Interstitial lung disease - Mushroom worker’s lung - *Pleurotus*

Introduction

Hypersensitivity pneumonitis (HP) is a underdiagnosed interstitial lung disease, with putative systematic manifestations, caused by excessive immunological response of the lung parenchyma to a variety of inhaled antigens, to which it is previously sensitized.1,2 These antigens include bacterial (mainly thermophilic actinomycoses, but also mycobacteria), fungal or animal proteins (e.g. pigeon breeder disease), as well as some chemical substances of low molecular weight (e.g. isocyanates). Exposure to these antigens, though most frequently occupational (e.g. farmers’ lung), may also be domestic or recreational (e.g. hot tub lung).3,5 Despite the insufficient epidemiological...
data, disease incidence in the UK is estimated at about 1 per 100,000 person-years, with a mortality rate three times higher than that of the general population.\(^6\) In the absence of widely accepted diagnostic criteria, HP often represents a diagnostic challenge, requiring a high level of clinical suspicion. Diagnosis is based on the identification of antigen exposure and the combination of clinical, imaging, laboratory (most importantly cytological analysis of bronchoalveolar lavage, BAL) and, less commonly, histological findings.\(^3,5\) The traditional classification into acute, subacute and chronic form is lately under scrutiny.\(^7,8\) In all cases, chronic HP is characterized by radiological and histological presence of lung fibrosis. Furthermore, its clinical features resemble those of idiopathic pulmonary fibrosis,\(^9-12\) thus the discrimination of the two clinical entities becomes frequently troublesome.\(^13-16\) Treatment is based on the, occasionally unfeasible, avoidance of exposure to the responsible antigenic factor, while systematic corticosteroids hasten recovery, without long-term benefit demonstrated in prospective clinical trials.\(^3,4,17\)

We report the case of a Pleurotus species mushroom grower diagnosed with hypersensitivity pneumonitis. To the best of our knowledge, this is the first case described in Greece.

**CASE PRESENTATION**

A 47 year old woman presented to the emergency department (ER) due to persistent productive cough with mucoid sputum, breathlessness in exertion, pleuritic chest pain and weight loss (~10kg) during the past three months.

The patient had an unremarkable medical history. She did not suffer from any known allergies, was an occasional smoker and had been working in a Pleurotus mushroom growing unit for the last six months.

The patient had been examined by a pneumonologist two months before presentation regarding the same symptoms. High resolution chest CT scan (HRCT) performed 40 days prior admission revealed patchy ground-glass opacities with symmetrical distribution, mainly in the upper lobes, and a small peripheral consolidation expanding to the right upper lobe, as well as residual fibrosis in both apaxes, without any other pathologic findings (Figure 1). The patient underwent bronchoscopy one month prior admission, in which patchy bronchial mucosal edema and purulent secretions were observed. Washing, BAL and transbronchial biopsies were conducted in the right middle lobe. BAL analysis demonstrated neutrophilia and eosinophilia, without lymphocytosis (Table 1), malignant cells or foamy macrophages, and negative D-galactomannan. Histological examination of the transbronchial biopsy specimens showed mild nonspecific chronic inflammatory infiltration. Bacteriological and fungal cultures were negative. At that time, other significant laboratory test results included an elevated CRP (25,80 mg/dl with a normal range of <6), slightly elevated erythrocyte sedimentation rate (ESR 59 mm/h) and low total immunoglobulin E value (IgE 44,8 IU/mL). Taking into account BAL analysis findings and after performing spirometry which revealed the presence of a mild restrictive pattern [FEV1 1,72 L (70% pred.), FVC 2,13 L (74% pred,) and FEVI/ FVC 80,8%], the patient was prescribed amoxycycline/clavulanate antibiotic treatment. Doxycycline was added to the antibiotic regimen seven days later, due to initial treatment failure. However, as symptoms still persisted, the patient eventually presented in ER. Off note, during the whole treatment period, she had continued to work at the same Pleurotus mushroom unit.

At the time of admission, the patient was afebrile, hemodynamically stable and mildly tachypneic (22 breaths per minute). Oxygen saturation was 96% on room air. Bilateral squeaks in upper and middle lung fields were present in auscultation, without any other remarkable physical examination findings or laboratory test results. Chest X ray revealed bilateral poorly defined small opacities, predominantly in the upper and middle lung fields (Figure 2). Autoantibodies test (RF, ANA, ACA, anti-dsDNA, anti-Scl-70, anti-CCP, anti-Jo1) and interferon-γ release assays (IGRAs) were negative. Bronchoscopy was repeated. In accordance to the first examination, bronchial mucosal inflammation with mucopurulent secretions in the upper lobes was found. Washing and BAL were, this time, performed at lingula. Quantitative BAL cultures, cytological examination of BAL and washing fluids for malignancy and β-Koch test results were negative. BAL differential cell count differed from that of the previous bronchoscopy (Table 1). Although predominant neutrophilia was again noticed, significant lymphocytosis was additionally observed, without accompanying eosinophilia. Regarding lymphocyte subpopulations, increased levels of CD8 T-cells and a corresponding low CD4/CD8 ratio were noted.

The patient was discharged at a stable clinical condition. Antibiotics were discontinued and she was instructed to abstain from work, until reevaluation on an outpatient basis.

The patient was reassessed six weeks later. She reported avoidance of exposure to mushrooms after discharge and
showed pronounced clinical improvement, with cough and chest pain resolution. Weight loss had ceased and dyspnea on exertion ameliorated. Lung function tests exhibited a rise in expiratory volumes [FEV1 1.89 L and FVC 2.25 L]. Finally, a new HRCT at that point, revealed a substantial decrease of the ground-glass opacities and a complete disappearance of the consolidation (Figure 3). The patient was advised for a permanent occupational change.

**DISCUSSION**

HP is a form of interstitial lung disease caused by inhalation of small (<5μm), mostly organic, but also inorganic particles, which provoke granulomatous inflammatory response in predisposed individuals, when reaching small airways and alveoli. The list of causative antigens is extensive, with fungi being frequently reported. In the HP group study, the largest until now published disease

**TABLE 1. BAL subpopulations 40 days before and during hospitalization.**

<table>
<thead>
<tr>
<th></th>
<th>1st Bronchoscopy</th>
<th>2nd Bronchoscopy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cell count</td>
<td>NA</td>
<td>118,25</td>
</tr>
<tr>
<td>(x 10⁴/mL)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Macrophages</td>
<td>28</td>
<td>24</td>
</tr>
<tr>
<td>Neutrophils</td>
<td>65</td>
<td>43</td>
</tr>
<tr>
<td>Lymphocytes</td>
<td>2</td>
<td>32</td>
</tr>
<tr>
<td>Eosinophils</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>CD4/CD8</td>
<td>NA</td>
<td>0.60</td>
</tr>
</tbody>
</table>

NA: not available.
cohort, involving 199 patients, fungi were recognized as the causative agent in 13% of cases. Exposure to *Penicillium* species is implicated in several types of HP, such as suberosis and peat moss worker’s lung, as well as certain cases of humidifier’s lung, while *Aspergillus* species is linked to malt worker’s lung, tobacco grower’s lung and occasionally farmer’s lung. *Trichosporon cutaneum* is considered responsible for the majority of cases of familial summer-type HP encountered in Japan. HP has been described in mushroom growers and is commonly referred to in literature as mushroom worker’s lung (MWL). Interestingly, the inciting antigenic factor varies depending on the species of cultivated mushroom. MWL was originally reported in growers of the *Agaricus* species, which continues to hold the largest share of global and local production, and was attributed to bacterial (and not fungal spores) inhalation, mainly of *Thermoactinomyces vulgaris* and *Micropolyspora faeni* originating from the compost used for cultivation. With the gradual worldwide expansion of other mushroom species production, such as *Pleurotus* and *Lentinus edodes*, MWL was also proved to develop in growers of the latter. However, in this case, pathogenesis is different and mainly due to inhalation of fungal spores (bradiospores) released in the air in large quantities during cultivation. The isolation of IgG antibodies against these spores from the sera of *Pleurotus* and *Lentinus* workers who developed MWL is indicative of the central role of fungi in the pathogenesis.

Our patient had been working in the production of *Pleurotus* mushroom species during the last six months, and presented respiratory symptoms approximately three months after the beginning of mushroom exposure. The duration of exposure before disease onset varies significantly and appears to be dependent on the nature of antigen, the intensity of exposure and host factors. In previous reports of MWL in *Agaricus* growers this latent period was approximately five years on average. In subsequent clinical series of MWL in *Pleurotus* workers, duration of engagement to cultivation prior symptom onset was occasionally not more than three months, as in the case of our patient.
There are no currently available specific tests or diagnostic criteria for HP. Diagnosis relies on the establishment of exposed to the potentially inciting antigen, clinical symptoms, HRCT pattern, BAL analysis, immunological testing and, frequently but not always, open lung biopsy. In our case, clinical suspicion of HP as the cause of the patient’s symptoms was based on the occupational exposure to mushrooms and the indicative HRCT pattern of patchy ground-glass opacities symmetrically distributed predominantly in the upper lobes. Typical HRCT findings of acute and subacute HP comprise ground-glass opacities, most often bilateral and symmetrical and/or patchy, centrilobular solid or, most commonly, ground-glass nodules and air trapping. Measurement of precipitins against Pleurotus spores is not available in our hospital. However, precipitins diagnostic value is limited by lack of standardization, low specificity (about 10% of asymptomatic farmers have positive precipitin testing) and significant number of false negative results in patients with HP. Repeat bronchoscopy and BAL were considered imperative for further diagnostic work-up. BAL analysis revealed significant lymphocytosis (>30%), with a low CD4/CD8 ratio, findings suggestive of HP diagnosis, although not conclusive. However, the constellation of exposure history, radiologic findings and BAL lymphocytosis was considered adequate for clinical diagnosis. It should be noted that according to the HP study group, which developed and subsequently validated a clinical prediction rule for the presence or absence of HP, the combination of bilateral ground-glass opacities or centrilobular nodules in HRCT and BAL lymphocytosis (≥30% for never or ex-smokers and ≥20% for active smokers) was adequate for HP diagnosis without further lung biopsy testing. Our patient met exactly these criteria.

The patient had been subjected to bronchoscopy one month prior admission. At that time, BAL presented neutrophilia and eosinophilia, but not lymphocytosis, and she had been prescribed empiric antibiotic treatment. While on antibiotics, she continued her occupation. Despite rendering HP as a less likely possibility, BAL neutrophilia does not rule out the diagnosis. In fact, early reports suggest that BAL differential leucocyte count in HP patients depends on the time interval since antigen exposure. Fournier and colleagues underwent ten HP patients to BAL both prior to and 24 hours as well as five or eight days after inhalation challenge. A dramatic increase of BAL neutrophils 24 hours after exposure resolving after 5 or 8 days was observed. It is noteworthy that in several patients of this cohort neutrophil predominance in BAL was found, as was also noted in our patient. Similar findings were reported by Drent and colleagues, who performed BAL in 59 HP patients at different time points after last exposure. Significant neutrophilia and eosinophilia were noted in BAL fluids extracted within 24 hours from antigen inhalation, with a tendency to return to control values at subsequent time points. Given that our patient kept working during diagnostic work, observed neutrophilia could be attributed to recent (<24 hours) contact with fungal spores. In our opinion, BAL lymphocytosis in the repeat procedure abolished the need for further testing. To the best of our knowledge, this is the first report of HP in mushroom worker in Greece.

The fundamental therapeutic intervention in HP treatment is elimination of exposure to the inciting antigen and in the majority of acute and subacute cases is sufficient for complete resolution of the disease. Systemic corticosteroids constitute the only accepted pharmacologic treatment. They are used in severe cases for recovery acceleration, without established long-term benefit. In the case of our patient, the slowly progressive nature of disease abolished the need for corticosteroids. Therapeutic approach was initially limited to an instruction for provisional work discontinuation and, following clinical, functional and imaging improvement, a permanent occupational change was recommended.

In conclusion, HP is a potentially treatable interstitial lung disease closely related to occupational and environmental exposures, as in the form of MWL presented in our patient. High level of clinical suspicion, early diagnosis and exposure elimination are key elements of the management of these patients.

COMPETING INTERESTS

All the authors declare that they do not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript.

REFERENCES